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Proceedings/ Summaries of Fourth Eastern Forage Improvement Conference

Beltsville Agricultural
Research Center
Beltsville, Maryland
July 7-9, 1981

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SYMPORIUM: MORE ON FORAGE QUALITY

Chairperson - James H. Elgin, Jr.

Introduction

The Fourth Eastern Forage Improvement Conference was held at the Beltsville Agricultural Research Center, Beltsville, Md., from July 7 through July 9, 1981. Drs. Stanley A. Ostazeski and James H. Elgin, Jr., were co-chairpersons for local arrangements. The formal program was arranged by the executive committee of EFIC.

The conference was opened by Chairperson James H. Elgin, Jr., on Tuesday at 1:30 p.m. Dr. Paul A. Putnam, Director, Beltsville Agricultural Research Center, welcomed participants.

The conference began with a symposium entitled "More on Forage Quality." Four papers were presented, which are reproduced here. Considerable discussion followed--a result of interest generated by the speakers and the quality of their presentations.

On Wednesday morning, July 8, participants reviewed current experiments of the alfalfa project at Beltsville. In the afternoon, they toured the facilities of the Forage Farm of the University of Maryland at Clarksville and W-L Research, Inc., at Highland, Md. The day's activities were concluded by a buffet dinner and fellowship in the Bioscience Building at the Beltsville Agricultural Research Center.

On Thursday morning, contributed papers were presented followed by the business meeting. The conference was adjourned at noon.

Differences in Quality of Tall Fescue Entries as Measured by Animal Studies

J. B. Powell and J. Bond¹

What is it about a grass that causes some farmers not to grow it on their farm, whereas others have exclusively seeded their pastures to this species. Some swear by it, others swear at it. Tall fescue (*Festuca arundinacea* Schreb.) is one of those grasses. Its value to farmers engaged in animal agriculture has been debated extensively for many years. The cow-calf enterprise of the Midsouth flourishes where this species excels. While other species of cool-season grasses have remained constant, increased some, or declined in the past 15 years nationally, total production of tall fescue seed has nearly tripled. Some of this increase in popularity is related to turf use, but most of the increase is attributed to pasture use. Even though tall fescue has excellent agronomic qualities for pasture and hay, animal performance has been erratic. Several animal health problems have been associated with this grass. This is the feature that makes it most controversial. I will mention only two health problems here because a complete review has been published (Bush, Boling, and Yates, 1979²).

Fescue foot, a lameness of animals grazing tall fescue, is one disorder that is readily recognized. Associated with it is rough hair coat, arched back, reduced rate of gain, and in advanced stages necrotic tail, ears, and feet. The disorder appears during late fall and winter during cold weather when animals are allowed to graze on accumulated foliage of tall fescue. Removing the animals from the forage corrects the problem. The toxic principle in the forage has never been rigorously defined, even though much is known about the toxic substance. Since management of the grass and the animals can largely eliminate this disorder, it is not the focus for the studies reported here.

The major animal disorder on which we focus is known as "summer syndrome" or "summer slump." It occurs during hot weather. The animals decrease grazing time, intake is lowered, gain is decreased and often becomes negative, and milk production drops off. Not all cattle in the same pasture show this response nor does it occur every grazing season on all varieties of tall fescue. The animals have an overwhelming urge to cool off. They lie in water and filth and spend much time around the water tank. They seek shade and avoid full sun.

Others observed that summer syndrome corresponds with high concentration of alkaloids in summer growing fescue and that perloline is a major component of numerous alkaloids in tall fescue.

¹ Director's Office and Ruminant Nutrition Laboratory, respectively, Beltsville Agricultural Research Center, Beltsville, Md. 20705.

² See References Cited, p. 3.

Perololine was seen to inhibit in vitro ruminal cellulose digestion, alter fatty acid production, and kill or inhibit microbes of the rumen. Since perloline content of breeding populations of tall fescue was found to be highly heritable, a method was devised to test the effects of perloline. By developing breeding lines having contrasting levels of high and low perloline and grazing them in pasture trials, distinct characteristics of animal behavior and performance should be revealed. R. C. Buckner at Lexington, Ky., developed such tall fescue lines.

The research undertaken at the Beltsville Agricultural Research Center (BARC) was to establish pastures for a 3-year grazing trial of the Buckner-developed lines. We considered that a broad cooperative approach involving many specialists and scientific disciplines could be applied to this problem. Buckner was supportive of this approach and furnished us with four entries--two with contrasting perloline levels [GL-306 (high perloline) and GL-307 (low perloline)], a newly released tall fescue called Kenhy, and a check, K-31.

The pasture design was presented to A. M. Decker of the University of Maryland, H. G. Matches of Columbia, Mo., and R. C. Buckner for their advice. D. J. Undersander of the University of Maryland (now at Texas Tech.) assisted us in laying out the plots and in managing the cattle and pastures the first year.

Agricultural engineer R. F. Dudley at BARC seeded the four entries. We used 2 replicates and 3 grazing rotations for a total of 24 pastures. Initially, 16 Holstein steers and 16 Angus steers were randomly assigned to replication and entry. Reserve animals were used in a put-and-take grazing design. All animals were weighed every 2 weeks through the grazing season. Visual scores of hair coat, health, condition, weight of salt blocks, measure of water intake, blood samples, behavior activity, and other measurements were taken. The forage was also sampled every 2 weeks prior to the animals entering a new pasture and after the animals were removed.

A short movie was presented that gave a visual image of the summer syndrome expression of animals grazing tall fescue.

The results were unexpected. The pasture having the low perloline line (GL-307) contained animals clearly showing summer syndrome symptoms when the environmental temperature was high. This line was expected to give better performance because of its low perloline content. J. Bond, who is the leader of this study, and I were perplexed. We communicated our finding over the telephone to Buckner in Kentucky, who with R. W. Hemken was conducting a test of the high and low perloline lines using fresh forage fed to dairy cows, and to J. O. Robbins at the Richard B. Russell Research Center, Athens, Ga., who had isolated loline alkaloids from fescue and was cooperating with C. W. Bacon and J. K. Porter, also at Athens, in studying the toxicity of Balansia and Epichloe fungi in grasses.

We sent freeze-dried forage samples to S. G. Yates, chemist, at the Northern Regional Research Center, Peoria, Ill. We dug up fresh plants from low and high perloline pastures as well as prepared large dry matter samples and sent them to J. O. Robbins for microbial and alkaloid analysis at the R. B. Russell Research Center in Georgia. We had the samples tested for sterols in our Tobacco Laboratory here at BARC and sent samples to R. A. Anderson of the University of Kentucky who tested them for phenols. Samples were sent to C. B. Smith at Penn State University for mineral analysis and to E. Cary of the U.S. Soil and Nutrition Laboratory at Cornell for selenium analysis. J. Velasco of the Seed Research Laboratory here tested the samples for aflatoxins. P. Lentz of our Mycology Laboratory examined these pastures for evidence of fruiting bodies of Balansia and S. A. Ostazewski of our Field Crops Laboratory searched for other fungal diseases. We talked to M. C. William of the Toxic Plant Center, Utah, on the nature of the problem. L. Daniels, University of Arkansas, used extracts from samples of our forage on pregnant rabbits. D. Kern, University of Maryland, fed hay from our plots to rabbits in heat chambers during balance studies.

H. Tyrrell and P. Moe of the Energy Laboratory here at BARC set up a study to test green-chopped forage daily for 6 weeks in controlled chambers. We visited the Middleburg, Va., Forage Research Farm of Virginia Polytechnic Institute and State University to view firsthand their pastures where animals on one Kenhy pasture had shown fescue toxicity, swollen feet, rough hair coat, and a generally unhealthy look from laying in water and filth. Our entomologists at BARC, R. Ratcliffe and P. Baker, netted insects in these pastures with the hope of picking up differential populations. We ran neutral-detergent fiber, acid-detergent fiber, lignin, and cellulose on all samples as well as in vitro digestibility dry matter determinations (K. Goering). D. J. Undersander of the University of Maryland analyzed for nonstructural carbohydrates. Our cereal quality laboratory (J. Wiser) looked at total protein and amino acids in the forage samples. We analyzed blood from the animals. We called staff meetings of all scientists at BARC interested in forage and forage-related research and exchanged views on the first results of the study.

R. F. Barnes, formerly staff scientist at Beltsville, called a Tall Fescue Workshop in Athens, Ga. We placed a contract with S. Wilson, Indiana University at Bloomington, to look into the synthesis of perloline and devise better tests for its quantification.

What is the significance of the results of the pasture test? Here, for the first time, summer syndrome could be induced in one tall fescue line and not in another. A clear and distinct behavioral reaction was induced in animals on the problem pastures. Heat stress was clearly a major factor to be related to decreased animal weight gain and in grazing behavior. This fact could explain the previously mentioned variability and erratic results. It also provided further suggestions for followup experiments. The animal

response found here was similar to the response noted by Hemken et al., 1979,³ on dry matter intake of flail-harvested fresh forage, milk production, and respiration rates of animals.

The second major break came when Shelly Yates at Peoria reported back on the samples sent to him in August 1977 from the problem pastures. He found perloline content of GL-306 (high) and GL-307 (low) as expected. However, GL-307 contained much more of the loline alkaloids (N-acetylloline and N-noracetylloline) than GL-306, which contained only a trace. J. Robbins in 1976 had reported these levels of loline alkaloids in samples we had sent to him. Thus, loline became suspect in the summer syndrome problem. A heat chamber experiment with animals fed the tall fescue, high and low in perloline, was the third major advance in understanding the problem. All our chambers were in use at BARC so we were unable to explore this avenue. Kentucky scientists were able to do these critical tests and confirmed the hypothesis that loline level in the forage was associated with summer syndrome. A near duplicate pasture study of the one at BARC was conducted in Kentucky. Differences were not so dramatic, but appearance of steers grazing GL-307 was similar to those observed at BARC. A fourth major advance came when Bacon, Porter, and Robbins found the endophyte fungus (*Epichloe typhina*) in samples of plants sent from Beltsville. Kentucky researchers also found the endophyte in their pastures. All pastures had some of the endophyte, but the GL-307 had a higher percentage. Thus, an association of loline alkaloids and the endophyte was established.

Work at Kentucky gave the next major advance in defining the nature of the problem. By controlling the endophyte fungus, loline also is controlled. By systemic fungicide treatment of plants (thereby excluding the fungus from the seed produced on the treated plants), an uncontaminated source of seed gave plants having low loline. Two approaches at Kentucky were taken at this point: (1) To produce contaminated and uncontaminated seed of the same line for pasture studies, and (2) to develop resistance lines that exclude the fungus.

Since 1976, we have gone full circle in the experimental process. New breeding materials are being readied by Buckner and grazing trials are planned for BARC and other locations. Observations have been extended to sheep reproduction. We have a much better understanding of a problem with tall fescue utilization that has limited its full value as a pasture species. During the 5 years of these tests, two events stand out that made this progress possible. First, two national tall fescue toxicosis conferences were held that put all scientists working on or interested in the problem together in a fact finding, current awareness, free exchange of idea format; secondly, information was communicated freely by telephone, letters, visits, and presentations. A large group

of scientists became aware of the problem and began to contribute in their discipline area.

The scientific method and people who make it work are alive and well. I have great confidence that we will continue to approach researchable problems of the complexity of this one in a multidisciplinary manner. This retrospective view of progress on quality improvement of tall fescue exemplifies the benefits and advances to be derived from a multidisciplinary approach and I urge its continuation.

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³ See footnote 2, p. 1.

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Application of Forage Quality Measurements to Plant Breeding //

C. C. Lowe¹

Both plant and animal scientists have contributed to developing the laboratory assay methods used to approximate animal feeding values of forages. Application of the methods to different uses, however, has presented some unique problems. Relatively slow progress has been made in adapting the methods for use in selection for quality in forage breeding programs.

An initial difficulty concerns definition of breeding objectives. Preferably they should be specific and quantifiable, i.e., yield of plant dry matter. Forage quality, in contrast, is vague and ambiguous referring to any or all the multiple factors involved in animal feeding values. Although consensus does exist on the importance of major forage quality constituents, there is less agreement on the relative significance of different assay measurements to infer animal performance. The breeder's choice of assay methods to use in selection depends on this interpretation, on method accuracy, and also on the involved mechanical difficulties and inherent costs for different assay procedures. With different species of diverse chemical composition used for forage and several types of animal utilization, it is logical that different assay methods and combination will be needed to best fit different breeding situations. Application knowledge will accumulate slowly.

Another initial dilemma in breeding for forage quality improvement lies in inherent quality differences between species. Genetic variance for quality criteria (and opportunity for breeding progress) is definitely greatest in those forage species on the lower end of the relative quality scale. A breeder must choose between better odds for improving a least-used species or the poorer odds of making a most-used species slightly more desirable. At Cornell, the second path had been followed in a decade of effort on improving forage quality in alfalfa. This crop is usually considered to rank highest among forages in average feeding value.

Our first consideration was the growth pattern of alfalfa in our environment. Usual management involves harvest of forage from three growth cycles each season; age of harvested forage from each cycle is about 6 weeks. Distribution of seasonal dry matter production under this sequence is about 45-30-25 percent for the successive harvests.

If digestible dry matter (DDM) and protein content are used as quality criteria, all the alfalfa grown from this type of production ranks very high to high in quality relative to other types of forage. Even so, substantial change in forage

quality of alfalfa occurs within each growth cycle. Average DDM content, for example, might change from 70 to 60 percent as each crop matures. Differences between genotypes or populations for quality criteria at any point in time within the growth cycle are much smaller than the total increment of change occurring in all genotypes within each growth cycle.

Recognizing this, a first step was to reduce the amount of quality change and maintain higher average quality for the season by shortening each growth cycle. Breeders were successful in doing this and increasing alfalfa productivity in the process by breeding types able to tolerate more intensive harvest. Usual cutting management was increased over time from two cuts per season to three and average quality was raised because of less mature forage on each date of harvest. Extensive studies showed the more productive growth types were definitely not lower in average quality on a season-long basis under this management. This destroyed some commonly held earlier theories about close association of alfalfa quality with plant growth habit and some easily observable morphological distinctions such as stem size.

Our next selection direction was based on plant fractions. With leaves known to be higher in average values for quality criteria than stems, we tried selection based on the ratio of leaves to stems for same aged forage and we also utilized a unique genetic trait that increases the leaflet number in alfalfa from the normal three to higher multiples--usually five or seven leaflets per leaf. In time we successfully produced varieties in which this multiple leaflet condition was expressed by about 95 percent of the plants in a variety population. Overall results of this selection for the multifoliate condition to performance has been a slight increase in average digestibility, a slight decrease in average protein content, a 10 percent loss in forage productivity, and indications of a possible drop in seed production potential. A very unique and desirable looking alfalfa was produced, but it has not been proved advantageous in feeding value and is at least questionably deficient in some important agronomic properties. Our efforts on using leaf-stem separation as a selection tool did improve on average forage quality, but this method is not very practical because of the labor requirements.

All studies of genetic variation in alfalfa for quality constituents using approximative assay procedures show the genetic ranges in current germplasms to be relatively narrow. The basic problem for alfalfa then is detection of small quality differences while restricted to using limited numbers of fairly expensive laboratory assays. This situation has determined the nature of our present quality selection. It is essential to maintain a current level of productivity and agronomic desirability during quality selection rather than to use extreme quality variants and then try to upgrade the agronomic properties. This rule probably applies rather generally to quality improvement in all forages.

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Our quality improvement program has also included development of diverse new germplasms from previously unused species. We are using these with recurrent selection but have no current assessment on whether this has increased genetic diversity for quality factors. A sequential selection system has evolved where we first reduce the source population in each cycle to about 5 percent by screening for agronomic traits. The survivors are then assayed for quality using the Udy dye binding method and acid-detergent fiber assay to reflect protein and digestibility. We have carried six source populations into a second selection cycle. We have definite evidence of breeding programs for both protein and fiber based on performance of synthetics derived from first cycle selections. We do not currently know how much we can alter the chemical composition of the crop without affecting agronomic properties and we have not yet evaluated the selection products in live animal feeding trials.

As we proceeded with quality selection within alfalfa, it became apparent that our major problem lay in forage sampling to reflect average differences between genotypes over a total growing season. This included two or three successive growth cycles with different environmental parameters and desirable constituents undergoing constant change within each cycle. This suggested a necessary minimum of sampling at appropriate points in time within several growth cycles. Additional requirements were suggested from studies of contributions to sampling error from location, and sample collection and preparation effects. Procedures have been modified by these studies. Accuracy for the various assay methods is a very minor consideration compared to plant sampling in use of these methods for forage breeding.

We are currently using assay procedures that best fit our particular objectives and resources. We are engaged in research that could lead to further refinements in sampling, and we are interested in any modification of assay procedures that could reduce assay costs and increase numbers of plants that can be screened for forage quality.

245 [CJ] How NIR Can Help in Measuring Forage Quality for Breeding and Utilization Programs //

J. S. Shenk

Infrared spectroscopy has been an important analytical tool for the chemist since commercial instruments were introduced in 1944. The first use of this technology was for qualitative analysis of liquids using infrared light transmitted through the substance. The near infrared (NIR) part of the spectra, although rich with information, was not used because of the complex mathematical computations needed to extract the information from the spectra. Reflectance rather than transmission measurements were needed to generate NIR spectra for solid agricultural products such as forage and grain. Successful quantification of the NIR spectra involves chemistry, electronics, and computer technology. Three companies--Dickey-John, Technicon, and Neotec--are currently manufacturing instruments for agricultural application.

Advantages of NIR

The major advantage of NIR reflectance is rapid analysis of multiple constituents. After the instrument is properly calibrated for a particular substance, such as hay or hay crop silage, analysis of successive samples requires only seconds per sample. It usually requires more time to open the sample container and place the sample in the holder than to do the analysis.

Other advantages are simplicity of sample preparation and ease of instrument operation. All that is required for sample preparation is grinding the sample. In addition, chemicals or reagents are not used, eliminating the associated safety hazards and pollution problems. The instrument can be operated by a relatively unskilled technician, using a very small workspace.

Disadvantages of NIR

The major disadvantage of NIR is instrument calibration. Representative samples with accurate wet chemistry data must be obtained to calibrate the instrument. This critical step has a large effect on the accuracy of predictions. The best way to overcome the problem is to have a State or Federal agency accumulate an official calibration set of samples and provide them to laboratories or private individuals for NIR analysis.

Plant Breeding With NIR

Plant breeders have been looking for a rapid analytical technique so that new varieties could be developed with higher quality as well as higher

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The author expresses his appreciation for the funds and personnel supplied by a cooperative project between the USDA-ARS and The Pennsylvania State University to conduct this research.

yield. NIR is the procedure to accomplish this breeding objective. Results from studies at Pennsylvania State University with orchardgrass are a case in point. Using NIR to analyze 1,500-2,000 samples per year, the analysis cost per sample was only 20 percent of the cost with wet chemistry.

NIR also provides a more accurate analysis. This is especially true for in vitro dry matter disappearance (IVDMD). Since the NIR instrument is calibrated with a small set of samples analyzed in a single batch, bias or nonrandom errors are eliminated. There is no satisfactory method of comparing the wet laboratory results with those predicted with NIR since comparative studies are no longer being conducted by this laboratory. However, estimates of narrow sense heritability for the quality traits protein, neutral-detergent fiber, and IVDMD range from 0.20 to 0.64 over more than 4 years of work and 7,000 analyses.

This technology is not being used by many forage breeders for a number of reasons. First and foremost is cost of instrumentation. If a forage breeder wants to analyze samples for multiple constituents, such as protein, fiber, digestibility, or moisture, the instrument and associated computer would cost between \$38,000 and \$75,000. Take as an example the equipment currently recommended for forage breeders that sells for \$46,000. If a breeder analyzed 2,000 samples for 4 constituents each year over a 5-year period, equipment costs alone would average about \$1.13 per analysis. Add to this part-time labor, calibration costs, instrument maintenance, and repair. The total cost would be approximately \$1.70 for each analysis or \$6.80 per sample.

The cost of obtaining this quality information with wet chemistry would be more than double the NIR cost per sample. In addition, the entire procedure is under the control of the plant breeder who only needs to use the instrument about 1 month each year. The technology really becomes cost effective if several breeders have access to the instrumentation. Currently NIR is being used to analyze forage in breeding programs in Pennsylvania, Minnesota, and New Mexico.

Prediction of Hay Quality

A number of studies have been conducted to test the prediction of hay quality with NIR. Hay grown under Pennsylvania conditions contains many different mixtures of grass and legume species harvested at different stages of maturity. The following tabulation shows the lowest NIR prediction errors published for hay. This represents the potential accuracy of the technology.

<u>Analysis</u>	<u>NIR standard errors of prediction</u>
Protein	0.66
Acid-detergent fiber	1.02
Neutral-detergent fiber	1.97
Lignin59
In vitro digestion	1.80
Dry matter35
Calcium16
Phosphorus04

These NIR prediction errors for hay were of the same general magnitude as the errors we have found among laboratories using the standard wet chemistry procedures. These results are possible with (1) a high quality scanning instrument, (2) careful control of sample processing, (3) proper selection of calibration samples, and (4) appropriate computer software to select wavelengths and develop prediction equations. These are the four keys to successful results with NIR.

Pennsylvania Hay Marketing Study

We attended three hay auctions in Pennsylvania to evaluate the nutrient variation in loads of hay, investigate sampling techniques, and become acquainted with the logistics of providing NIR analyses under local marketing conditions. Hay samples were taken at the market and returned to the university for analysis by both wet chemistry and NIR. An average load of hay consisted of approximately 100 (50 to 60 lb) bales. The variation of quality parameters within a load was about twice the variation within a single bale (table 1).

Table 1. Errors of sampling individual bales of hay and variation in hay loads (standard deviations)

Source of variation	Protein	Acid-detergent fiber (ADF)	Neutral-detergent fiber (NDF)
Individual bales	0.65	1.41	1.68
Hay loads	1.54	2.41	4.17

The limiting factor in sampling was the time required to take the sample. Using the Penn State hay sampler (a 24- by 3/4-inch core sampler attached to a 1/2-inch battery-powered electric drill), at least 2 minutes were required to collect a composite sample from five bales chosen at random from the load. If this sample were analyzed by current wet chemistry procedures, the standard error of the load determinations based on one composite sample would be +0.8 percent for protein, +1.2 percent for ADF, and +2.1 percent for NDF. If an NIR instrument was used, the errors would be +1.0 percent for protein, +1.9 percent for ADF, and +2.6 percent for NDF. This higher error for NIR would be easily offset by the speed of analysis and availability of the information.

A comparison was made of the price paid for hay and its quality as determined by NIR analysis.

The correlation of this relationship over the 385 loads of hay was +0.63. Only 40 percent of the variation in the price paid for hay could be attributed to hay quality. We concluded that the NIR procedure would be fast enough to operate under working conditions at the market and accurate enough for decisionmaking. Furthermore, the potential savings to the buyers and sellers made a self-supporting analysis system seem economically feasible. Since the NIR analysis for this study was not conducted at the hay markets, it was decided to develop a mobile NIR analytical system to interact directly with the farmers at these hay auctions.

The NIR Van

The NIR instrument and computer were mounted in a standard size van along with a grinder to prepare the sample for analysis. This analytical system was taken to the hay market auction at Belleville, Pa., for testing. Hay loads were sampled, analyzed, and the results posted on the load before the sale. Analyses displayed included crude protein, total digestible nutrients, and hay grade determined by the computer from the equations developed by the American Forage and Grassland Council.

The response by the farmers to this analysis and grading program changed throughout the 5 weeks of the study. In the first 2 weeks, little attention was paid to the posted information beyond expressions of curiosity. By the third week, farmers bringing hay from the same field and same harvest were finding the analyses to be very repeatable.

This began to build confidence in the analyses. Buyers were beginning to study the analyses as well as the hay on the truck. Some farmers selling low quality hay began to refuse analysis and buyers showed little interest in purchasing their hay. During the fourth week, some sellers with high quality hay wanted us to continue the service, and by the fifth week, more farmers were requesting that we continue the service. At this point the program was discontinued because of other planned research activities.

Our accomplishments during the 5-week period can be summarized as follows: First, we learned how to make the van system function efficiently. Second, we demonstrated that NIR analysis of hay under marketing conditions can be accomplished without disrupting the sale in any way. Third, we found that NIR analysis does not give the buyer or the seller an advantage in the market but does provide a basis on which the auction can function more equitably. Fourth, we found a great lack of knowledge among farmers about hay quality and how to use this information in their feeding program.

Expanding the Van Concept

Having developed a satisfactory mobile hay analysis system, we proceeded to devise a method for analyzing high moisture samples of corn silage, hay crop silage, high moisture corn, and green chop. Samples were dried 5-7 minutes in a microwave oven giving a rapid turnaround time for

high moisture analysis. NIR prediction errors were not enlarged because of microwave drying if the instrument was calibrated using microwave dried samples. A computer program for ration formulation was then added to complete the mobile analytical and ration formulation system.

In Pennsylvania we estimate that one mobile NIR system operated by a full-time person trained in analysis and ration formulation could be a profitable enterprise for the hay producers, instrument operators, and dairymen if it supports 3,000 dairy cows.

The items required for this mobile system were a grinder-vacuum device for grinding samples and cleaning sample holders, a microwave oven to dry high moisture samples, an electronic balance to determine "as is" moisture in high moisture samples, a scanning filter Nectec 51 NIR instrument, DEC computer, terminals, air conditioner-heater combination for winter and summer operation, a self-contained electrical power system and customized computer software for instrument control, nutrient prediction, and ration formulation. The equipment was available "off-the-shelf" from various suppliers, and the computer software was a Pennsylvania State University/U.S. Department of Agriculture development. The van complete is available from P.A.G.E., Allentown, Pa.

Conclusion

Both mobile and stationary NIR systems are now available. The best choice of instrumentation depends on its application. The important point is that both types of systems have been developed and are commercially available if needed. We believe that NIR will be a useful technology for forage breeders as well as for the entire hay and dairy industry of this country.

H. K. Goering¹

Preservation of forage for winter feeding is required in most areas of the United States. Daily animal production is a function of intake, digestibility, and energetic efficiency or feed conversion. Animal production per acre is also a function of recovery of available forage from the field and storage. Recovery of forage from the field is maximized by direct cutting and storage. Direct-cut forage must be dehydrated or ensiled.

Much of the work at Beltsville has been with direct cutting and ensiling of hay crops. The problems encountered with direct-cut silage are low lactic acid, high pH, high butyric acid, low intakes, high soluble nitrogen, and high seepage losses. Formic acid addition as a 90 percent solution at 0.5 percent of wet weight successfully overcomes all the problems with the exception of controlling protein degradation, which can be controlled using a combination of formic acid and formaldehyde. Heifers have gained a kilogram a day when fed direct-cut alfalfa silage preserved with a formic acid:formaldehyde mixture. Storage recovery is improved about 5 percent by using these additives or formic acid by itself. The technique of direct cutting and storage has opened new potential in evaluating high quality forages in utilization experiments; immediate acidification and storage result in a predictable and consistent product in contrast to wilting and haymaking, which introduce many uncontrollable factors before storage. Alfalfa and orchardgrass were direct cut at the same digestibility and ensiled with formic acid:formaldehyde mixtures. The intake, digestibility, growth rate, and energetic efficiency of Holstein steers and heifers were determined. Alfalfa-fed cattle grew faster, consumed more dry matter, and utilized metabolizable energy more efficiently. Body fill as a proportion of body weight was 17.9 percent for alfalfa-fed steers compared with 23.2 percent for orchardgrass-fed steers.

Ammonia has been used as an additive to all types of forage in recent years. Most work has been with the ensiling of the whole corn plant. The research on ammonia shows it can reduce protein degradation during ensiling, increase silage stability, improve the silage fermentation, improve energy recovery, and reduce incidence of heating after removal from the silo. Ammonia is also an economical source of nonprotein nitrogen for cattle and sheep, and performance data indicate that it is equal to urea. When ammonia is added at 4 percent or more of the forage, it will also improve the fiber digestibility of low quality forages. Use of more legumes, along with chemical preservation techniques, gives new options for the present and future in raising, handling, storage, and utilization of winter feed for ruminants.

CONTRIBUTED PAPERS

Session Chairperson - Jack E. Winch

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Sampling Variation and Plant Compositional Changes in Alfalfa Selected for Nutritional Quality¹James Coors¹

Sixteen alfalfa populations were evaluated for forage quality on a total season basis using a modified Udy dye-binding procedure (percent transmittance (T)), 48-hour *in vitro* rumen digestibility, and cell wall composition. Three of the populations were developed by intercrossing parental clones selected for agronomic performance, low acid-detergent fiber (ADF), and high percent T. These combinations were compared with corresponding genic populations developed from clones selected only on agronomic characteristics.

Two of the three populations derived from nutritional quality selection had higher percent T and lower ADF values than their unselected counterparts in each of three harvests. These improvements were associated with higher digestibility, lower neutral-detergent fiber, and lower lignin values.

Number of samples required per solid-seeded plot (3.9 by 1.0 m) and the number of replications needed to detect specified differences in percent T were calculated. For percent T, bulk samples formed by combining equal dry weights of multiple samples from a plot gave values equal to the means of the individual samples combined.

Population rankings based on percent T values remained stable over harvests. Ranks of several populations, however, dramatically changed from harvest to harvest for other quality parameters.

Digestibility and percent T values were highly correlated within individual harvests. The relationship nearly equals that between digestibility and ADF.

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Protein Quality as Determined by Physicochemical Properties of Forage Fractions

M. L. Fishman and R. A. Barford¹

A comprehensive method has been developed to extract and fractionate forage by differences in structure, solubility, and molecular size. Fractions have been characterized by nitrogen content, amino composition, and various parameters obtained from automated gel filtration, which include molecular weight distribution, changes in solubility, and changes in contamination by endogenous aromatic compounds. The methodology has been applied to various species of grasses to measure changes in protein brought about by environmental factors. The results from the studies will be reviewed.

Voluntary Intake and Digestibility of Wilted Grass Silages Prepared Under Humid Conditions of Maritime Canada

P. Narasimhalu, K. A. Winter, and H. T. Kunelius¹

First-cut Trader meadow fescue (MF), Tardus orchardgrass (OG), Norlea perennial ryegrass (PRG), Lemtal Italian ryegrass (IRG), and quackgrass (QG1), as well as second-cut quackgrass (QG2), were made into wilted silages and fed, each to six wether lambs, in two 24-day periods to measure the voluntary intake and apparent digestibility at 90 percent ad libitum intake. IRG was wetter, more acidic, and contained higher total volatile fatty acid concentration than the other silages. Voluntary intakes were not different among the first-cut silages but were considerably lower for the QG2 silage ($P>0.05$). Dry matter digestibility was highest for PRG or IRG and decreased in sequence for OG or QG1 and QG2 ($P>0.01$). Digestible dry matter intake of QG2 was only 75 percent of the QG1 mainly because of decreased intake. In vitro dry matter disappearance (IVDMD) by the rumen inoculum-pepsin method ranked the silages in descending order as follows: PRG, MF, IRG, QG1, OG, and QG2, whereas with the acid-pepsin solubility (APDMD) test, the ranking order was almost the same except QG2 rated higher than QG1 and OG. IVDMD rather than APDMD closely depicted the ranking order of in vivo digestibilities of the silages.

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Potential for Improved Nitrogen Fixation in
Alfalfa //

J. H. Stangarone,¹ A. M. Decker,² and J. O. Legg¹

The ability to improve legume nitrogen fixation could offer a means of biologically increasing forage protein and soil nitrogen. Plants previously selected from four cultivars of alfalfa (*Medicago sativa*) were designated as having the potential to fix high or low amounts of nitrogen (N). The major objectives were to determine whether differences in nitrogen fixation existed between plants designated as high- and low-fixers and to observe seasonal changes in nitrogen fixation. Cuttings of selected plants representing both high and low fixation rates were then made to increase numbers and to insure identical genetic traits. These cuttings were grown under field conditions on a 6.1- by 6.1-m plot, which contained ¹⁵N-labeled organic matter. Data were then collected on dry weight, percent N, and total mg N; percent N fixed and total mg fixed N were obtained by using isotopic dilution approaches. Orchardgrass (*Dactylis glomerata*) was grown among the alfalfa plants and served as the nonfixing control.

In general, no significant differences were observed among designated high- and low-fixing plants for percent nitrogen or percentage of fixed N in the plant. However, there were significant differences in dry weight, total mg N, and total mg N fixed among high and low entries with the first almost always exceeding the last. These trends were generally consistent throughout the harvest year.

Fertilization of No-Till Forages for Maximum Yield on Hill Lands //

A. M. Decker

The Northeast has many thousands of acres, capable of producing high yields of quality forage, that are currently idle or producing far below potential. No-tillage forage establishment offers an environmentally safe, economical way to improve the productivity of these hill lands, which are too steep for conventional tillage.

A study was undertaken to compare forage productivity of no-till forages on level fields with those on slopes too steep for conventional tillage. The objective of the study was to compare forage yields of four sod-seeded mixtures at two fertilization rates on a level Chester soil and a nearby steep, eroded Manor soil. A randomized complete block experiment with four replications on each soil site was used in this study.

Weed problems associated with the spring no-till seeding delayed data collection until 1 year after seeding. Extreme drought during the third year resulted in severe stand losses and early termination of the study.

The data clearly demonstrate that surface application of lime and fertilizer can result in significant yield increases of no-till forages established on steep hill land soils. Yields can be comparable with those obtained on level soils that have been considered potentially more productive. During the first 2 years after seeding, red clover appeared comparable to alfalfa in terms of yields. Orchardgrass seeded with alfalfa resulted in increased forage yields and fewer weeds.

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Comparison of In Vitro and Traditional Propagation Methods in Red Clover and Birdsfoot Trefoil

D. T. Tomes, C. T. Campbell, and B. Orshinsky¹

Shoot tip culture has been used to store and propagate several hundred genotypes of birdsfoot trefoil for up to 4 years with routine subculture at yearly intervals. Storage at 4° C is less time and space consuming than traditional storage in growthroom or field plantings plus offering much higher survival rates. Recently, the shoot tip culture procedures have been adapted for red clover. Similar responses were noted in red clover and trefoil in that benzyladenine (BA) produces more shoots, and no hormones result in fewer shoots but more roots. Similar success with storage at 4° was also noted for red clover. Red clover requires higher levels of BA (2.0 mg per liter compared with 0.05 mg per liter) to achieve higher levels of shoot multiplication. Crown buds are required for efficient establishment of a shoot tip culture in red clover, whereas axillary buds can be used effectively in birdsfoot trefoil. Further extension of long-term storage of birdsfoot trefoil through cryopreservation of shoot tip and callus cultures has so far shown limited success.

In vitro and traditional cutting methods of propagation were compared for agronomic performance in eight genotypes of red clover of genetically diverse origin. Genotypes were significantly different for all traits measured. Seed and forage yield and regrowth were similar for in vitro and traditional cuttings. In vitro propagation resulted in significantly fewer flowers, better winter survival, and a better spring vigor rating. Plant vigor at first flower and forage yield in the year after establishment were also significantly higher for in vitro propagation. This method appears to have merit for propagation in red clover.

Genetic stability of in vitro propagation is being investigated in red clover and birdsfoot trefoil. Preliminary results in red clover suggest that the in vitro methods are as stable genetically as traditional propagation methods. Similar experiments are in progress with birdsfoot trefoil in field and growthroom experiments.

Use of Standard Clones and Hypodermic Inoculations for Race Identification of *Colletotrichum trifolii*

S. A. Ostazeski and J. H. Elgin, Jr.¹

We have described the use of a hypodermic inoculation technique to nondestructively determine the resistance to *Colletotrichum trifolii*, race 1 and race 2, of alfalfa plants in breeding and genetic studies. During the development of this technique, we isolated alfalfa clones with the combination of resistant (R) and susceptible (S) reactions to race 1 and race 2, respectively: RR, clones 1 and 52; RS, clones 16 and 54; SS, clones 4 and 31. With these clones we have verified the classification to race of all *C. trifolii* isolates in our collection. These same clones can be used to classify field-collected specimens of anthracnose. Stems bearing anthracnose lesions are incubated in petri dish moist chambers, or in plastic bags containing moistened paper towels, in a refrigerator at about 10 to 15° C. After about 3 days' incubation, lesions are usually sporulating profusely. A sporulating lesion from a given collection site is cut from the stem, placed in a test tube containing 2 to 6 ml of "Tween 20" solution (1 drop of Tween 20 in 500 ml distilled water), and shaken vigorously to dislodge and suspend conidia. About 1 ml of the suspension is drawn from the tube into a hypodermic syringe. Sites on succulent stems of each of the six clones are marked and identified with the collection site data by hanging marking tags at the node below the internode to be inoculated. Inoculations are made with 23-27 gage needles at minimal plunger pressure. Readings can be made about 10 to 14 days after inoculation.

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BUSINESS MEETING

Fourth Eastern Forage Improvement Conference

Beltsville Agricultural Research Center
Beltsville, Md. 20705
July 9, 1981

The meeting was called to order at 11:10 a.m. by the chairperson, Dr. James H. Elgin, Jr. The minutes of the Third Eastern Forage Improvement Conference (EFIC) held at Carlton University, Ottawa, Ontario, July 10-12, 1979, were read and approved.

Old Business

In regard to the feasibility of joint meetings with other organizations, the chairperson reported that nothing had been done to change the ongoing meeting arrangements. The conference members apparently value the unique merits of small group meetings such as we presently experience.

Nothing has been done on getting ISS numbers for past and future issues of the EFIC proceedings. The present secretary will, in cooperation with Dr. Jack Winch, arrange for such assignment as soon as certain communications problems have been corrected.

New Business

Dr. Winch reported that he has had requests for past issues of EFIC proceedings. He further stated that he is the custodian of some of the past issues, as are Drs. Elgin and A. Morris Decker. He elaborated on the merit and need of a central archives and for a permanent secretary for EFIC. Considerable discussion ensued, after which Dr. Winch moved that the alfalfa project at Beltsville, Md., be designated the permanent site for the archives of EFIC. Action on the assignment of a permanent secretary for EFIC was postponed for further study.

Dr. Carl Lowe inquired of the chairperson what effect competition from other, possibly more prestigious, meetings had on the final decision to hold our regular EFIC conference. Discussion followed with the conclusion that, despite competition, the small size of EFIC has no effect on the quality of its meetings nor the enthusiasm of its participants.

The chairperson reminded the EFIC that its incoming chairperson, Dr. Winch, would be the representative to the National Alfalfa Improvement Conference and that its next meeting in 1982 was to be held at the University of California, Davis.

Discussion developed on the availability of proceedings from the Central and Western Alfalfa Improvement Conferences. The chairperson pointed out that, in addition to our own membership, our proceedings are sent only to the executives of the other conferences. He further stated that he would endeavor to get our mailing list to the other conferences and thus probably expedite receipt of other conference reports.

The following committees were appointed by the chairperson:

<u>Nominations</u>	<u>Resolutions</u>
D. T. Tomes C. C. Lowe, Chairperson	A. M. Decker R. Michaud, Chairperson

The nominations committee proposed the following slate to hold office until the next regular meeting:

Chairperson - J. E. Winch, University of Guelph, Guelph, Ontario
Vice chairperson - S. A. Ostazeski, USDA, Beltsville, Maryland
Secretary - N. Lawson, MacDonald College, Ste. Anne de Bellevue, Quebec

No others were nominated from the floor, and it was moved, seconded, and approved by the conferees that the slate be accepted.

The resolutions committee submitted the following resolution:

Inasmuch as the Beltsville Agricultural Research Center and the University of Maryland served as hosts for the Fourth Eastern Forage Improvement Conference, and whereas Drs. James H. Elgin, Jr., Stanley A. Ostazeski, and A. Morris Decker did an outstanding job of organizing and handling the conference and the field tour, be it resolved that we, the participants of the conference, extend our sincere thanks to these individuals for their efforts in making our stay at Beltsville productive and enjoyable.

Inasmuch as W-L Research, Inc., kindly received us on the field tour, be it further resolved that we, the participants of the conference, extend our sincere thanks to Drs. Angus A. Hanson, David F. Beard, Joseph H. Graham, and other staff members for their excellent hospitality and congratulate them for their excellent program of developing and testing alfalfa varieties. We move for the adoption of these resolutions.

The chairperson reminded the membership that at our 1979 meeting we had been invited to MacDonald College, Ste. Anne de Bellevue, Quebec, for our 1983 meeting. Dr. Lowe invited the group to Cornell University, Ithaca, N.Y., for the 1985 meeting.

As a result of the transfer of power, Dr. Winch, the incoming chairperson, adjourned the meeting.

Stanley A. Ostazeski
Secretary, Fourth Eastern Forage Improvement Conference

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